(12) UK Patent Application (19) GB (11) 2 080 108 A

(21) Application No 8115494
(22) Date of filing
20 May 1981
(30) Priority data
(31) 803054
803121
(32) 22 May 1980
26 May 1980
(33) South Africa (ZA)
(43) Application published
3 Feb 1982
(51) INT CL³ A61K 31/575
(52) Domestic classification
A5B 180 190 248 24Y 281 28Y 382 38Y 39X H
A2B 311 321 611 613 770 ECD
(56) Documents cited
GB 1553540
GB 949256
(58) Field of search
A5B
(71) Applicant
Louis Harold Krut
55 Alexandra Avenue
Craighall
Johannesburg
Republic of South Africa
(72) Inventor
Louis Harold Krut

(54) Pharmaceutical compositions containing oxidised cholesterol

(57) Oxidation products of cholesterol which act synergistically with phospholipids to promote solubility of cholesterol in vitro also promote solubilization and clearance of crystalline cholesterol deposited in tissue in humans. The oxidation products of cholesterol are formed by synthetic processes or are allowed to form spontaneously from cholesterol in foods of animal origin by subjecting such foodstuffs to natural or artificial ultra-violet light.

GB 2 080 108 A

stuff is solid, at least the larger are preferably exposed to the ultra-violet light. The foodstuff is also preferably cut fairly thin, and is preferably not more than 4 cm, preferably not more than 2 cm thick.

The invention is illustrated by refernce to the following non-limiting examples.

EXAMPLE 1

Oxydation products comprising 7-hydroxy cholesterol, 7β-hydroxy cholesterol, 7-keto cholesterol and 6-cholesten-3β,5-diol were made by dispersing cholesterol (obtained from Merch AG Germany) in ethyl alcohol in an alkaline solution of sodium stearate and bubbling oxygen through at 85 C for 5 hours. The products comprised approximately 9,8% of each of the 7 and 7β-hdroxy cholesterol, about 7,7% of the 7-keto cholesterol and about 2,2% of 6-cholesten-3β,5-diol. The mixture used was used in the proportions in which it was formed.

A supersaturated solution of cholesterol in sunflower seed oil was formed at 90 C, and the solution poured into a number of separate test tubes which were then held in a water bath at 25 C.

Phosphatidylcholine from egg yolk was added to one of the test tubes. Phosphatidylcholine is present in the human body and is known to micellise cholesterol. Under the above conditions the phosphatidylcholine did not significantly increase the solubility of cholesterol.

In another test tube, a small amount of oxidation products formed above was added in addition to the phosphatidylcholine. The two products acted synergistically and increased the solubility of cholesterol in supersaturated solution enormously.

EXAMPLE 2

The previous example was repeated in a supersaturated aqueous solution of cholesterol. With cholesterol and phosphatidylcholine present in equal amounts by mass, only some of the cholesterol is readily dispersed in the water.

When the oxidation products of cholesterol are added to the cholesterol and phosphatidylcholine system there was observed a ready dispersal of all the cholesterol as well as its oxidation products.

EXAMPLE 3

The preceding examples were repeated using spontaneously generated oxidation products of cholesterol prepared as will be described below. I found that the same results occurred as with the artificially produced oxidation products of cholesterol.

EXAMPLE 4

A 500 ml amount of milk at room temperature and in containers with the surface area of about 80 square centimetres located at about 10cm from a light source and was subjected to stirring for about 30 minutes.

The light source was a B–100A Black Ray Lamp made by Ultraviolet Products Inc of San Gabriel, California which at 15 inches has an intensity of 7000 uW cm with a peak emission of 365 nanometers.

An amount of oxidation products of cholesterol were formed equivalent to the amount that would be formed if the milk had been allowed to stand in daylight for three days. The palatability of the milk was unaffected. The milk was drunk by a human subject without any harmful effects being noticeable. Further amounts of milk made in this way was consumed daily (one litre per day) by the human subject for about ten weeks with no harmful effects noted.

EXAMPLE 5

Slices of meat about 2cm thick was exposed to ultraviolet light from the abovementioned light source from a distance of about 30cm. Both sides of the meat were so exposed for a period of 30 minutes. I found that oxidation products of cholesterol were formed in the same amounts as in meat standing in daylight for three days. The meat did not decompose and its palatability was unaffected. This meat and meat similarly treated was also consumed by a human subject over the same time period as the milk treated as described for a period ten weeks with no harmful effects being noted.

EXAMPLE 6

Four types of tablets were made up containing the following active agents:— Tablets A, cholesterol alone; Tablets B, cholesterol and oxysterols; Tablet C, cholesterol and phosphatidylcholine; and Tablets D, cholesterol, oxysterols and phosphatidylcholine. The oxysterols were formed of the compounds and in the proportions mentioned above. The tablets were made up in various masses between 45 mg to 78 mg.

The tablets were implanted under the skin of twenty four rats each having a body mass of between 250 g and 300 g. Two tablets were implanted subcutaneously on the belly of each rat at about 40 mm distance as follows:—

Group 1	Tablets A and B
Group 2	Tablets A and C
Group 3	Tablets A and D
Group 4	Tablets B and C
Group 5	Tablets B and D and
Group 6	Tablets C and D

I found that I was able to recover Tablets A wholly from the rats after four weeks. There had been no change in these tablets which had been covered with a fibrous tissue that

had grown over them. The tablets B and D had disappeared totally. The tablets C were recovered with a 25% reduction in mass.

The experiment was repeated with another twenty four rats for a period of four weeks with substantially identical results.

The experiment was repeated again with twenty four rats but on this occassion they were examined after four months. Once more tablets B and D had disappeared. No change in tablets C from the condition extracted from the rats at four weeks was noted. The tablets A had no observeable change whatsoever from thhe original form in which they had been implanted. None of the rats showed any ill effects from any of the tablets.

I further repeated the experiment with tablets A and B only with smaller numbers of rats and examined the tablets seven to ten days after implantation. I found that tablets B had undergone liquifaction to the consistency of thin cream and had begun to be transported away. As I expected tablets A had undergone no change.

I deduced that the phospholipids delivered to the site of the tablets naturally acted synergistically with the oxidation products of cholesterol to solubilise the cholesterol which is the reason why the tablets containing cholesterol and the oxidation products of cholesterol dissolved as did those which contained cholesterol, phosphatidylcholine and the oxidation products of cholesterol.

From the above observations I have deduced that any or all of the oxidation products of cholesterol would be beneficial to man by preventing the abnormal deposition and/or promoting the clearance of cholesterol so deposited in tissues such as arteries. As is well known the abnormal deposition of cholesterol in the arterial wall leads to atherosclerosis which is directly or indirectly reponsible for causing diseases such as heart attacks and strokes.

The invention is not limited to the precise details hereinbefore described. The spirit and scope of the invention is to be determined solely from the appended claims.

CLAIMS

1. A pharmaceutical composition comprising an oxidation product of cholesterol together with a pharmaceutically acceptable carrier.

2. A composition as claimed in claim 1 further including a phospholipid.

3. A composition as claimed in claim 2 wherein the phospholipid is phosphatidycholine.

4. A composition as claimed in claim 1, 2 or 3 comprising a plurality of the oxidation products of cholesterol.

5. A composition as claimed in any one of the preceding claims in which the oxidation product of cholesterol comprises between about 250 mg and 1000 mg.

6. A method of treating a foodstuff comprising subjecting the foodstuff to intense artificial ultra-violet light.

7. A method as claimed in claim 6 in which the foodstuff that is treated contains cholesterol.

8. A method as claimed in claim 6 or 7 in which the foodstuff is a liquid comprising causing the liquid to flow past a source of ultraviolet light.

9. A method as claimed in claim 8 wherein the liquid is caused to flow in a thin film.

10. A method as claimed in any one of claims 6 to 9 wherein the foodstuff is a dairy product.

11. A foodstuff when treated by the method claimed in any one of the preceding claims 6 to 10.

Printed for Her Majesty's Stationery Office
by Burgess & Son (Abingdon) Ltd.—1982.
Published at The Patent Office, 25 Southampton Buildings,
London, WC2A 1AY, from which copies may be obtained.